Claims

1. A method of determining the amount or presence of antibodies to an immunogen in a blood sample comprising at least the steps of:

detecting the amount or presence of plasma antibodies or parts thereof and lymphocyte antibodies or parts thereof in said sample, wherein said plasma and lymphocyte antibodies are detected together or in separate assays and determination of their combined amount or presence determines the amount or presence of antibodies to said immunogen.

2. A method as claimed in claim 1 wherein:

the lymphocytes of said blood sample are disrupted whereby to release antibodies or parts thereof associated with said lymphocytes; and

the amount or presence of said lymphocyte antibodies or parts thereof released from the lymphocytes and the amount or presence of plasma antibodies are detected.

3. A method as claimed in claim 1 or 2 additionally comprising the step of:

isolating a plasma containing sample from said blood sample; and

isolating a lymphocyte containing sample from said blood sample.

4. A method as claimed in any one of claims 1 to 3 wherein said whole blood sample is divided into two portions for the preparation of a lymphocyte containing sample and a plasma containing sample.

5. A method as claimed in any one of claims 1 to 3 wherein a single whole blood aliquot is used to prepare a lymphocyte containing sample and a plasma containing sample.

6. A method as claimed in any one of claims 3 to 5 wherein said lymphocyte containing sample and/or said plasma containing sample is a purified preparation.

7. A method as claimed in any one of claims 3 to 6 wherein said plasma containing sample and said lymphocyte containing sample are recombined prior to disrupting the lymphocytes.

8. A method as claimed in any one of claims 3 to 6 wherein said plasma containing sample and said lymphocyte containing sample are recombined after disrupting the lymphocytes and prior to said detection step.

9. A method as claimed in claim 7 or 8 wherein the ratio of said plasma containing sample and lymphocyte containing sample is between 1:0.4 to 1:4, preferably 1:1.

10. A method as claimed in any one of claims 1 to 9 wherein said plasma and lymphocyte antibodies are detected in a single assay.

11. A method as claimed in any one of claims 1 to 6 wherein said plasma and lymphocyte antibodies are detected in separate assays.

12. A method as claimed in any one of claims 1 to 11 wherein said blood sample is a mammalian, preferably human blood sample.

13. A method as claimed in any one of claims 1 to 12 wherein said blood sample is less than 1 ml.

14. A method as claimed in any one of claims 1 to 13 wherein red blood cells are removed from said blood sample.

15. A method as claimed in any one of claims 1 to 14 wherein non-B lymphocytes are removed from said blood sample.

16. A method as claimed in any one of claims 1 to 15 wherein said immunogen results from infection or vaccination.

17. A method as claimed in any one of claims 1 to 16 wherein said immunogen is a bacterial or viral antigen, preferably an antigen from the group selected from antigens from Herpes Simplex Virus Cytomegalovirus, human immunodeficiency virus (HIV), a hepatitis virus, Epstein-Barr virus, Aphthovirus, Toxoplasma, tuberculosis, syphilis and chlamydia.

18. A method as claimed in any one of claims 1 to 17 wherein the lymphocytes are disrupted by using chemical disruption buffers (preferably containing detergent) or physical disruption means.

19. A method as claimed in any one of claims 1 to 18 wherein said antibodies or parts thereof are detected by contacting said antibodies or parts thereof with one or more antigens or antibodies which recognise said antibodies or parts thereof, preferably carried on a solid phase.

20. A method as claimed in any one of claims 1 to 19 wherein the released antibodies are detected by means of a solid phase binding assay.

21. A method as claimed in claim 19 or 20 wherein said solid phase carries one or more antigens (solid phase antigens) recognised by the antibodies or parts thereof to be detected (target antibodies).

22. A method as claimed in claim 19 or 20 wherein said solid phase carries one or more antibodies (solid phase

antibodies) recognised by the antibodies or parts thereof to be detected (target antibodies).

23. A method as claimed in any one of claims 1 to 22 wherein the detection step is performed by immunoassay, preferably ELISA.

24. A method as claimed in any one of claims 19 to 23 wherein one or more antigens, recognised by the target antibodies immobilised on said solid phase, are contacted with said solid phase.

25. A method as claimed in any one of claims 19 to 23 wherein one or more antibodies, recognised by the target antibodies immobilised on said solid phase, are contacted with said solid phase.

26. A method as claimed in any one of claims 1 to 25 wherein said detection step takes place in solution.

27. A method as claimed in any one of claims 1 to 26 wherein a soluble substrate is used for the detection step and yields a spectrophotometrically detectable signal.

28. A method as claimed in any one of claims 1 to 27 wherein a negative control is used.

29. A method as claimed in any one of claims 19 to 28 wherein multiple solid phases are employed, each bearing a different antigen or antibody which recognises a different target antibody.

30. A method as claimed in any one of claims 1 to 29 wherein the blood sample for use in the method and/or the lymphocyte containing sample and/or the plasma containing sample is stored at a temperature of 4°C or less before the lymphocytes are disrupted.

31. A method as claimed in claim 30 wherein the blood sample for use in the method is obtained from whole blood and said whole blood is frozen with 5-15% DMSO before the lymphocytes are disrupted.

32. A method as claimed in any one of claims 1 to 31 wherein the lymphocytes in said sample are not incubated under conditions to allow production and/or secretion of antibodies prior to said method.

33. A method as claimed in any one of claims 1 to 32 wherein multiple samples are tested simultaneously or sequentially.

34. The method as claimed in any one of claims 1 to 33 for use in identifying an animal infected by an immunogen, preferably HIV infected patients.

35. The method as claimed in claim 34 wherein infection of said animal by said immunogen, preferably HIV, occurs less than 10 days before the blood sample is obtained from said animal.

36. The use of the method as defined in any one of claims 1 to 35 in high throughput screening.

37. The use as claimed in claim 36 wherein said screening is of blood bank blood samples.

38. The method or use as defined in any one of claims 1 to 37 for determining the suitability of a sample for inter-individual transplantation or transfusion.

39. The use of the method as defined in any one of claims 1 to 35 or use as defined in claim 36 or 37 for diagnosing or monitoring infection of a human or non-human animal or a part of said animal by an immunogen, preferably a bacterium or virus, and the presence or extent of

infection by said immunogen, preferably by said bacterium or virus, is determined by reference to appropriate control and/or reference samples.